

Claims

We claim:

1. A method for purifying substantially pure and undegraded RNA from biological material comprising RNA, comprising the steps of:
 - (a) mixing said biological material, with an RNA Binding Solution buffered at a pH of greater than about 7, said RNA Binding Solution comprising an RNA-complexing salt, wherein said RNA Binding Solution is free of a strong chaotropic substance to form a mixture of biological material in RNA Binding Solution;
 - (b) contacting said mixture to a non-silica solid support such that nucleic acids comprising substantially undegraded RNA in said mixture preferentially bind to said solid support;
 - (c) washing said solid support with an RNA wash solution to remove biological materials other than bound nucleic acids comprising substantially undegraded RNA; and
 - (d) preferentially eluting the bound substantially undegraded RNA from said solid support with an RNA elution solution in order to obtain substantially pure and undegraded RNA.
2. The method of claim 1, wherein the biological material is selected from the group consisting of crude and partially purified mixtures of nucleic acids.
3. The method of claim 1, wherein the biological material is selected from the group consisting of eukaryotic cells, prokaryotic cells, microbial cells, bacterial cells, plant

cells, mycoplasma, protozoa, bacteria, fungi, viruses, yeasts, rickettsia and homogenates thereof.

4. The method of claim 1, wherein the biological material is selected from the group consisting of whole blood, bone marrow, blood spots, blood serum, blood plasma, buffy coat preparations, saliva, cerebrospinal fluid, and solid animal tissues.
5. The method of claim 1, wherein the biological material is selected from the group consisting of feces, urine, tears, and sweat.
6. The method of claim 1, wherein the biological material is selected from the group consisting of environmental samples taken from air, water, sediment and soil.
7. The method of claim 1, wherein the non-silica solid support comprises components selected from a group consisting of cellulose, cellulose acetate, nitrocellulose, nylon, polyester, polyethersulfone, polyolefin, polyvinylidene fluoride, and combinations thereof.
8. The method of claim 1, wherein the non-silica solid support comprises a polyester.
9. The method of claim 1, wherein the non-silica solid support comprises combinations of polyesters.
10. The method of claim 1, wherein the non-silica solid support is contained in a vessel, wherein the vessel is selected from a group consisting of centrifuge tubes, spin tubes, syringes, cartridges, chambers, multiple-well plates, test tubes, and combinations thereof.
11. The method of claim 1, wherein the strong chaotropic substance is selected from the group consisting of guanidinium salts and urea.
12. The method of claim 1, wherein the substantially pure and undegraded RNA is total RNA selected from the group consisting of messenger RNA, transfer RNA, ribosomal RNA

and viral RNA, and combinations thereof.

13. The method of claim 1, wherein the RNA-complexing salt is an alkali-metal salt.
14. The method of claim 13, wherein the alkali-metal salt is chosen from the group consisting of sodium, potassium, lithium, cesium, and rubidium salts.
15. The method of claim 13, wherein the alkali-metal salt is a lithium salt.
16. The method of claim 13, wherein the alkali-metal salt is lithium chloride
17. The method of claim 13, wherein the alkali-metal salt is present at a concentration greater than about 4 M.
18. The method of claim 13, wherein the alkali-metal salt is present at a concentration of between 4 – 10 M.
19. The method of claim 1, wherein the RNA Binding Solution optionally comprises a chelating agent.
20. The method of claim 19, wherein the chelating agent is selected from the group consisting of EDTA and CDTA.
21. A method for purifying substantially pure and undegraded RNA from biological material comprising RNA, comprising the steps of:
 - (a) mixing said biological material with an RNA Lysing Solution buffered at a pH of greater than about 7, said RNA Lysing Solution comprising an amphiphilic reagent, and an RNA complexing salt, wherein said RNA Lysing Solution is free of a strong chaotropic substance;
 - (b) lysing said biological material with said RNA Lysing Solution to form a lysate comprising nucleic acids comprising substantially undegraded RNA and non-nucleic acid biological matter;

- (c) contacting said lysate to an immobilized non-silica solid support such that said nucleic acids comprising substantially undegraded RNA in said lysate preferentially bind to said solid support;
 - (d) washing said solid support with an RNA wash solution to remove non-nucleic acid biological matter; and
 - (e) preferentially eluting the bound substantially undegraded RNA from said solid support with an RNA elution solution in order to obtain substantially pure and undegraded RNA.
22. The method of claim 21, wherein the biological material is selected from the group consisting of crude and partially purified mixtures of nucleic acids.
23. The method of claim 21, wherein the biological material is selected from the group consisting of eukaryotic cells, prokaryotic cells, microbial cells, bacterial cells, plant cells, mycoplasma, protozoa, bacteria, fungi, viruses, yeasts, rickettsia and homogenates thereof.
24. The method of claim 21, wherein the biological material is selected from the group consisting of whole blood, bone marrow, blood spots, blood serum, blood plasma, buffy coat preparations, saliva, cerebrospinal fluid, and solid animal tissues.
25. The method of claim 21, wherein the biological material is selected from the group consisting of feces, urine, tears, and sweat.
26. The method of claim 21, wherein the biological material is selected from the group consisting of environmental samples taken from air, water, sediment and soil.
27. The method of claim 21, wherein the non-silica solid support comprises components selected from a group consisting of cellulose, cellulose acetate, nitrocellulose, nylon,

polyester, polyethersulfone, polyolefin, polyvinylidene fluoride, and combinations thereof.

28. The method of claim 21, wherein the non-silica solid support comprises a polyester.
29. The method of claim 21, wherein the immobilized non-silica solid support comprises combinations of polyesters.
30. The method of claim 21, wherein the solid support is contained in a vessel, wherein the vessel is selected from a group consisting of centrifuge tubes, spin tubes, syringes, cartridges, chambers, multiple-well plates, test tubes, and combinations thereof.
31. The method of claim 21, wherein the strong chaotropic substance is selected from the group consisting of guanidinium salts and urea.
32. The method of claim 21, wherein the substantially pure and undegraded RNA is total RNA selected from the group consisting of messenger RNA, transfer RNA, ribosomal RNA and viral RNA, and combinations thereof.
33. The method of claim 21, wherein the RNA-complexing salt is an alkali-metal salt.
34. The method of claim 33, wherein the alkali-metal salt is chosen from the group consisting of sodium, potassium, lithium, cesium, and rubidium salts.
35. The method of claim 33, wherein the alkali-metal salt is a lithium salt.
36. The method of claim 33, wherein the alkali-metal salt is lithium chloride
37. The method of claim 33, wherein the alkali-metal salt is present at a concentration greater than about 4 M.
38. The method of claim 33, wherein the alkali-metal salt is present at a concentration of between 4 – 10 M.
39. The method of claim 20, wherein the amphiphilic reagent is a detergent.

40. The method of claim 39, wherein the detergent is a non-ionic detergent
41. The method of claim 40, wherein the nonionic detergent is selected from the group consisting of tweens, tritons, noniodets, and tergitols.
42. The method of claim 21, wherein the RNA Binding Solution optionally comprises a chelating agent.
43. The method of claim 42, wherein the chelating agent is selected from the group consisting of EDTA and CDTA.
44. A method for purifying substantially pure and undegraded RNA from biological material, comprising the steps of:
 - (a) contacting a biological material containing RNA, with a solid support pre-treated with an RNA Binding Solution buffered at a pH of greater than about 7 such that the RNA Binding Solution is bound to the solid support, said RNA Binding Solution comprising an RNA-complexing salt, wherein the RNA Binding Solution is free of a strong chaotropic substance;
 - (b) contacting said biological material to said solid support such that nucleic acids comprising substantially undegraded RNA preferentially bind to said solid support;
 - (c) washing said solid support with an RNA wash solution to remove biological materials other than bound nucleic acids comprising substantially undegraded RNA; and
 - (d) preferentially eluting the bound substantially undegraded RNA from said solid support with an RNA elution solution in order to obtain substantially pure and undegraded RNA.

45. A method for purifying substantially pure and undegraded RNA from biological material, comprising the steps of:

- (a) contacting a biological material containing RNA, with a solid support pre-treated with an RNA Lysing Solution buffered at a pH of greater than about 7 such that the RNA Lysing Solution is bound to the solid support, said RNA Lysing Solution comprising an amphiphillic reagent and an RNA-complexing salt, wherein said RNA Lysing Solution is free of a strong chaotropic substance;
- (b) contacting said biological material to said solid support in order to release nucleic acids comprising substantially undegraded RNA and non-nucleic acid biological matter causing nucleic acids comprising substantially undegraded RNA to preferentially bind to said solid support;
- (c) washing said solid support with an RNA wash solution to remove biological materials other than bound nucleic acids comprising undegraded RNA; and
- (d) preferentially eluting the bound undegraded RNA from said solid support with an RNA elution solution in order to obtain substantially pure and undegraded RNA.